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L1 0 S BACTERIA? (P) (POLYA (W) POLYMSERASE?)
L2 0 S BACTERI? (P) (POLYA (W) POLYMSERASE?)
L3 0 S PROKARYOT? (P) (POLYA (W) POLYMSERASE?)

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TI Further studies on the isolation and properties of polyriboadenylate polymerase from *Escherichia coli* PR7 (RNase I-, pnp).

AU Ramanarayanan M; Srinivasan P R

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AB Polyriboadenylate polymerase was isolated from *Escherichia coli* PR7 (RNase I-, pnp) in good yield and high purity. The enzyme catalyzes the polymerization of ATP and ADP. These polymerizations show an initial lag which can be removed by the addition of poly(A). However, poly(A)

does not function as a primer. UDP and CDP can also serve as substrates but with decreased efficiency. The polymerization of CDP is enhanced by the presence of an oligonucleotide which again does not function as a primer. Polymerization of [gamma-32P]ATP or [beta-32P]ADP result in products with no radioactivity. The product formed from [alpha-32P]ATP on hydrolysis with alkali yields labeled pAp and 2',3'-AMP; thus the enzyme synthesizes poly(A) chains de novo. During the polymerization of ATP, no burst of

free ADP can be detected and the time course of phosphate release from ATP and ADP follows very closely the kinetics of polymerization. dATP and dADP

are effective inhibitors of poly(A) synthesis from either ATP or ADP. Sulfhydryl reagents inhibit only the polymerization of ATP and the inhibition is fully reversed by dithiothreitol. However, the enzyme can

be protected from sulfhydryl reagents by preincubation with either ATP or

ADP in the absence of Mg²⁺ which is required for polymerization. Studies

using acrylamide gel electrophoresis indicate that the polymerization activity with either ATP or nucleoside diphosphates resides in the same protein. The enzyme catalyzes the following exchanges: 32Pi into ADP, 32Pi into ATP, and [14C] ADP into ATP in the presence of phosphate. While the

enzyme catalyzes the phosphorolysis of its own product, (pAp-(Ap)nA), it fails

to cleave the dephosphorylated product, (Ap(Ap)nA), or ribosomal RNA or tRNA in the presence of inorganic phosphate. The differences and similarities between **poly(A) polymerase** and polynucleotide phosphorylase are discussed. Based on the 32P exchange studies and other properties of **poly(A) polymerase**, a plausible mechanism for its action is proposed.

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